## REMARKS/ARGUMENTS

In an Office Action mailed on December 1, 2004 in the above-identified application, Claim 1-15 are currently pending. Claims 1 and 5-10 are withdrawn from consideration as being drawn to a nonelected invention. Claims 2-4 and 11-15 were rejected. Specifically, the Office Action has substantively rejected Claim 2 under 35 U.S.C. §101; Claims 2-4, and 12-15 under 35 U.S.C. §112, second paragraph (indefiniteness); Claims 2-4, and 11-13, and 15 under 35 U.S.C. §112, first paragraph (written description); Claims 2-4 and 12-15 under 35 U.S.C. §112, second paragraph (enablement) and Claims 2-4 and 11-15 under 35 U.S.C. §102(b). Each issue raised by the Examiner is considered separately below. In light of the amendments above, and the arguments below, reconsideration is respectfully requested.

Election/Restrictions

Claims 1 and 6-10 stand withdrawn from further consideration notwithstanding the applicants' request for complete reconsideration of the restriction requirement. Briefly, in a previous response to a requirement for restriction filed on September 7, 2004, applicants elected with traverse Group IV, drawn to a nucleic acid encoding SEQ ID NO:4, encompassing Claims 2-4 and 11-15. Based on that response, the Examiner revised the restriction and rejoined Groups III and IV into new Group II drawn to polynucleotide encoding SEQ ID NO:2, SEQ ID NO:1; SEQ ID NO:4, and SEQ ID NO:3 (i.e., Claims 2-4 and 11-15). Applicants acknowledge such rejoinder and thank the Examiner for having reconsidered the original restriction requirement. Although applicants now elect revised Group II, we also wish to reserve the right to file a divisional application drawn to the non-elected Claims 1, and 6-10 and/or to rejoin process Claims 6-10 once the product claims have been found allowable.

Claim Objections

Claims 11-14 are objected to as allegedly drawn to multiple inventions. In response, applicants amend independent Claim 11 as shown herein above to reflect the election.

Claim Rejections – 35 U.S.C. §101

Claim 2 stands rejected under 35 U.S.C. 101 because it is directed to non-statutory subject matter. In response, applicants amend Claim 2 to include the term "isolated" before "nucleic acid," at the suggestion of the Examiner.

Claim Rejections – 35 U.S.C. §112, second paragraph

Claims 2-4, and 12-15 stand rejected under 35 U.S.C. 112, second paragraph, as being indefinite. Specifically, the Examiner asserts that Claims 2 and 15 recite "under moderately stringent hybridization conditions" but that the metes and bounds are not clear. In response, applicants amend Claim 2 and 11 to recite the requisite hybridization conditions. Support for this amendment can be found, for example, at page 6, paragraph [00030] of the specification. Accordingly, Claim 15 is cancelled.

Similarly, the Examiner asserts that Claims 12 and 13 recite "a predetermined level of expression" but that the metes and bounds are not clear. In response, applicants amend Claims 12 and 13 to indicate that the predetermined level of expression for the positive and negative controls are quantitatively determined. Support for this amendment can be found or example, at page 3, paragraph [00019]; page 5, paragraph [00027]; page 8, paragraph [00036] and page 9, paragraph [00043].

The Examiner also rejected dependent Claim 14, drawn to kit comprising a polypeptide selected from the group consisting of SEQ ID NO:2 and 4, because the base Claim 11 does not include peptides as one of the recited components. In response, Claim 11 is amended to include the polypeptide sequences of Claim 14. Accordingly, Claim 14 is cancelled.

Claim Rejections – 35 U.S.C. §112, first paragraph

Claims 2-4, and 11-13, and 15 stand rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement. Specifically, the Examiner asserts that Claims 2-4, 11-13 are

> "interpreted as drawn to a genus of nucleic acid molecules with various degrees of variations i.e. 80% identity to SEQ ID NO:1, or 3, and hybridizes under moderately stringent hybridization conditions to the nucleic acid molecules with 80% identity to SEQ ID NO:1. (Claims 2-4), hybridizes to a nucleic acid that encodes polypeptide of SEQ ID NO:2, and 4 (Claims 11-13)."

In response, for clarity purposes applicants submit that independent Claim 2 is amended to recite suitable stringent hybridization conditions. Similarly, Claim 11 is amended to include the language of Claim 15 and specific hybridization conditions. Accordingly, Claim 15 is cancelled.

Furthermore, applicants submit that the specification clearly recites identifying characteristics of the claimed genus (i.e., the nucleic acid molecules) in several locations. At paragraph [0009], the specification provides that "the polypeptide-encoding polynucleotide

sequence has at least about 85% nucleotide sequence identity to the coding sequence of SEQ ID NO:1 or SEQ ID NO:3 (using the NCBI Blast 2 comparison protocol)..." At paragraph [00025], the specification also provides detailed structural information showing that the murine CRG-L1 (SEQ ID NO:1) encodes a polypeptide of about 275 amino acids and has seven putative transmembrane domains which correspond to amino acids 33-53, 62-82, 91-111, 123-143, 146-166, 174-194, and 212-232 of SEQ ID NO:2. The mouse sequence (SEQ ID NO:1) was also found to be related to sequences in *C. elegans* and *D. melanogaster* which suggest a conserved function for the polynucleotide (see paragraph [00038].)

The specification also discloses that sequences from a single mouse clone were arranged to form coding sequence 1-825 (plus a stop codon) of SEQ ID NO:3 (human), corresponding to a polynucleotide sequence from bases 35 to 862 of SEQ ID NO:1 from mice. Further, it is noted that such a sequence can encode a protein in humans that corresponds to the protein of SEQ ID NO:2. The putative human cDNA is 87% identical to the mouse sequence. If the putative human cDNA is translated, the resulting amino acid sequence is 91% similar to the corresponding portion of the mouse amino acid sequence, using the Lipman-Pearson protein alignment with a gap penalty of 4 and gap length penalty of 12 (see paragraph [00040].) All of this structural information about the claimed sequences serves to support the notion that sufficient identifying structural characteristics have been provided in the specification.

Claims 2-4, and 11-13, and 15 stand rejected under 35 U.S.C. 112, first paragraph, for lack of enablement for nucleic acid molecules beyond nucleic acids SEQ ID NO: 1 and 3, and nucleic acids encoding SEQ ID NO:2 and 4. Specifically, the Examiner asserts that similar sequences of unknown species encompassed for use as cancer markers are still unpredictable and that no working examples or guidance have been provided to allow a person of ordinary skill in the art to make and use nucleic acid species similar to SEQ ID NO: 1 or 3.

In response, applicants submit that indeed applicants have disclosed representative examples as well as statements showing that the disclosed representatives are applicable to the genus as a whole. Representative examples of the genus showing conserved nucleic acid and polypeptide sequences throughout evolution from *C. elegans* and *D. melanogaster* to murine and human were disclosed at paragraphs [00038] –[00040] of the specification.

Furthermore, based on the state of the art at the time of filing, using the specification, one of ordinary skill in the art would have been able to identify similar polynucleotide sequences that show differential expression in liver tumor cells as compared to normal liver tissue cells and used them as markers for identifying liver tumors. Specifically, the

specification disclosed that gene expression differences between liver tumors and regenerating liver could be determined using "Representational Differences Analysis" or RDA of cDNA (see Lisitsyn et al., Science 259:946 (1993) and Hubank et al., Nucleic Acid Research 22:5640-5648 (1994)). Next, it was disclosed that the presence and level of differentially regulated protein could be discerned using antibodies directed to epitopes on the protein using well known methods. Likewise gene expression in a liver tumor and in regenerating liver tissue can be measured using widely known methods and disclosed conditions for hybridizing nucleic acids (see paragraphs [00028] - [00030].)

In addition, the specification provides a detailed working example of how to make and use the claimed polynucleotide and polypeptide sequences (see paragraphs [00032] - [00044]. The specification provides a skilled artisan with the ability to assess using conventional methods the expression levels of the claimed polynucleotide and array of tissues and more specifically to monitor the expression of the gene in human liver tumors as compared to normal human liver tissue. Likewise, antibodies directed to a portion of the human protein can be produced and used as diagnostic agents for assessing protein levels in various human tissues including liver tumors, see paragraphs [00042] and [00043].

For these reasons, it is believed that the Examiner has not met the burden of showing a lack of enablement. Rather, the Applicants have presented evidence that shows the application as filed does teach a skilled artisan how to make and use the claimed invention without undue experimentation using available tools. Reconsideration and withdrawal of these rejections are respectfully requested.

Claim Rejections – 35 U.S.C. §102

Claims 2-4, 11, 13, and 15 stand rejected under 35 U.S.C. 102(b) as being anticipated by Bonaldo et al., (1996, Genome Research, vol. 6, pages 791-806). Applicants believe that the Examiner's rejection is flawed. It appears that the Examiner has selectively chosen pieces of Bonaldo et al., to formulate a rejection, but in doing so mischaracterizes the publication as well as the claimed invention.

In a lengthy (16 page) article, Bonaldo et al., discloses four different methods that were developed and used to generate normalized cDNA libraries from human, mouse, rat and parasites. In addition, it discloses the construction and preliminary characterization of a subtracted liver/spleen library (INFLS-SI) that resulted from the elimination (or reduction of representation) of ~5000 INFLS-IMAGE clones from the INFLS library. The Examiner,

however, points to the *NotI*-tag-$(dT)_{18}$ and other poly(dT) primers for priming the first strand of cDNA synthesis (see page 801, right column under the heading "Construction of Directionally Cloned cDNA Libraries"). The Examiner also points to Figure 6 which shows the sequence of the pT7T3-Pac polylinker and flanking sequences. Furthermore, the Examiner points to the "Abstract" asserting that it teaches extracts from fetal spleen or fetal liver. Based on this characterization, the Examiner concludes that the claimed nucleic acid reads on the poly(dT) primers of the art, given that SEQ ID NO:1 has polyA tails (about 20 A's) at the 3' untranslated region. Applicants respectfully disagree with this line of reasoning.

In response, applicants submit that the claimed nucleic acids of the invention are patentably distinguishable from the Bonaldo paper. Specifically, the paper does not teach or suggest how to make the claimed nucleic acids and polypeptides whose expression is deregulated in liver tumor cells from human and non-human animals, relative to the expression in regenerating liver tissue. Furthermore, by reading the article as a whole or the specific passages cited by the Examiner, one skilled in the art would not be able to determine how to use the claimed invention so as to monitor the expression of the claimed gene in human liver tumors as compared to normal human liver tissue. However, in an effort to expedite prosecution on the merits and for clarification purposes, applicants amend Claims 2 and 11 to recite the coding sequences of SEQ ID NOs:1 and 3. Support for such an amendment can be found throughout the specification, namely page 2, paragraph [0008]; page 5, paragraph [00029]; page 7, paragraph [00038]; and page 8, paragraph [00040]. Accordingly, applicants submit that the disclosure of Bonaldo et al., does not anticipate the claims of the present invention.

Claims 2, 11-13, and 15 stand rejected under 35 U.S.C. 102(b) as being anticipated by Wu et al., (April 12, 1996, Biochim Biophys Acta. Vol. 1315, issue No. 3, pages 169-75). Again, applicants believe that the Examiner's rejection is flawed.

Specifically, the Examiner asserts that Wu et al., at page 170 left column teach "oligo(dT) cellulose (Boehringer, Almere, The Netherlands)" that is used to isolate poly(A) RNA, and "Oligo(dT)-*Not1* (Invitrogen, San Diego, CA)" that is used to prime the first strand of cDNA synthesis. The Examiner further asserts that the claimed nucleic acid reads on either the oligo(dT) cellulose or "Oligo(dT)-Not1.

In response, applicants submit that Wu et al., reports the identification of 36 up- and down-regulated cDNAs from HCC and normal liver, using the substraction enhanced display technique. Wu et al., does not disclose the claimed nucleic acids or polypeptides. By simply

disclosing that RNA from liver was purified by "oligo(dT) cellulose (Boehringer, Almere, The Netherlands)" affinity chromatography and that "Oligo(dT)-Not1 (Invitrogen, San Diego, CA)" was used to prime the first strand of cDNA synthesis does not directly or indirectly lead to the disclosure of the claimed polynucleotide and polypeptide sequences. However, as indicated hereinabove, for clarification purposes, applicants have amended Claims 2 and 11 to recite the coding sequences of SEQ ID NOs: 1 and 3. Support for this amendment is found throughout the specification. Accordingly, applicants submit that the disclosure of Wu et al., does not anticipate the claims of the present invention.

In view of the above claim amendments and remarks, the application is now believed to be in condition for allowance. Applicants respectfully request that a timely Notice of Allowance be issued in this case.

A petition for a one month extension of time accompanies this response so that the response will be deemed to have been timely filed. Please charge the extension fee to Deposit Account No. 17-0055. If any extension of time is required in any subsequent response, please consider this to be a petition for the appropriate extension and a request to charge the petition fee to Deposit Account No. 17-0055. No other fee is believed to be due in connection with this response. However, if any fee is due in this or any subsequent response, please charge the fee to the same Deposit Account No. 17-0055.

Respectfully submitted,

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